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Outbreak of methicillin-resistant *Staphylococcus pseudintermedius* in a litter of puppies: evidence of vertical perinatal and horizontal transmission

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Abstract:

Objectives: Methicillin-resistant *Staphylococcus pseudintermedius* (MRSP) has recently emerged as an important animal health problem. In this study we describe the epidemiology of a MRSP outbreak in a litter of Boxer puppies. All puppies (n=12) were born healthy but showed signs of severe septicaemia during the first week and eventually died 10 days after birth. Pathological examination revealed multifocal haemorrhagic lesions in various organs.

Methods: Standard PCR tests were performed with DNA extracted from organs to identify *Brucella canis* and common neonatal viral pathogens such as Canine Herpesvirus and Canine Parvovirus type 1. Pure cultures of β -haemolytic staphylococci were isolated from all organs (liver, spleen, kidneys and blood taken directly from the heart). In the following weeks, staphylococci were also isolated from the bitch, the puppies' father, and the two owners, giving a total of 16 isolates. All isolates were identified as *S. pseudintermedius* by restriction fragment length polymorphism (RFLP) of the *pta* gene and were shown to be methicillin-resistant by PCR detection of *mecA*. Routine antimicrobial susceptibility testing was performed by the microbroth dilution using Sensititre. *Sma*I-pulsed field gel electrophoresis (PFGE) and *spa* typing was done on five isolates originating from blood taken directly from the heart of one puppy, the vaginal cavity of the bitch, and the nasal cavities of the father and the two owners. Staphylococcal Cassette Chromosome *mec* (SCC*mec*) was typed using M-PCR 1 and 2 as described by Kondo et al. (2007).

Results: None of the samples were positive for Canine Herpesvirus, Canine Parvovirus type 1 and *B. canis*. All staphylococcal isolates were confirmed to be MRSP and displayed the same resistance profile. In addition to β -lactam resistance, the isolates were resistant to fluoroquinolones (MIC >2 μ g/mL), chloramphenicol (MIC >16 μ g/mL), clindamycin (MIC >2 μ g/mL), gentamicin (MIC >8 μ g/mL) and tetracycline (MIC >8 μ g/mL). Vancomycin, teicoplanin, rifampicin and amikacin were the only antibiotics tested displaying *in vitro* efficacy. PFGE analysis revealed indistinguishable band patterns, indicating that the bitch, the father and the two owners were carriers of the same MRSP strain causing death of the puppies. All isolates harboured SCC*mec* type III and belonged to *spa* type t02, which is the most common European MRSP clone.

Conclusions:

1. This study shows that MRSP can be a cause of fatal neonatal outbreaks in dogs.
2. The fact that the same clone was isolated from the vaginal mucosa of the bitch strongly indicates vertical perinatal transmission of MRSP.
3. Isolation of the same MRSP clone from the two owners suggests that these bacteria can be horizontally transferred to humans living in close contact with dogs and that dog owners may act as vehicles for MRSP